

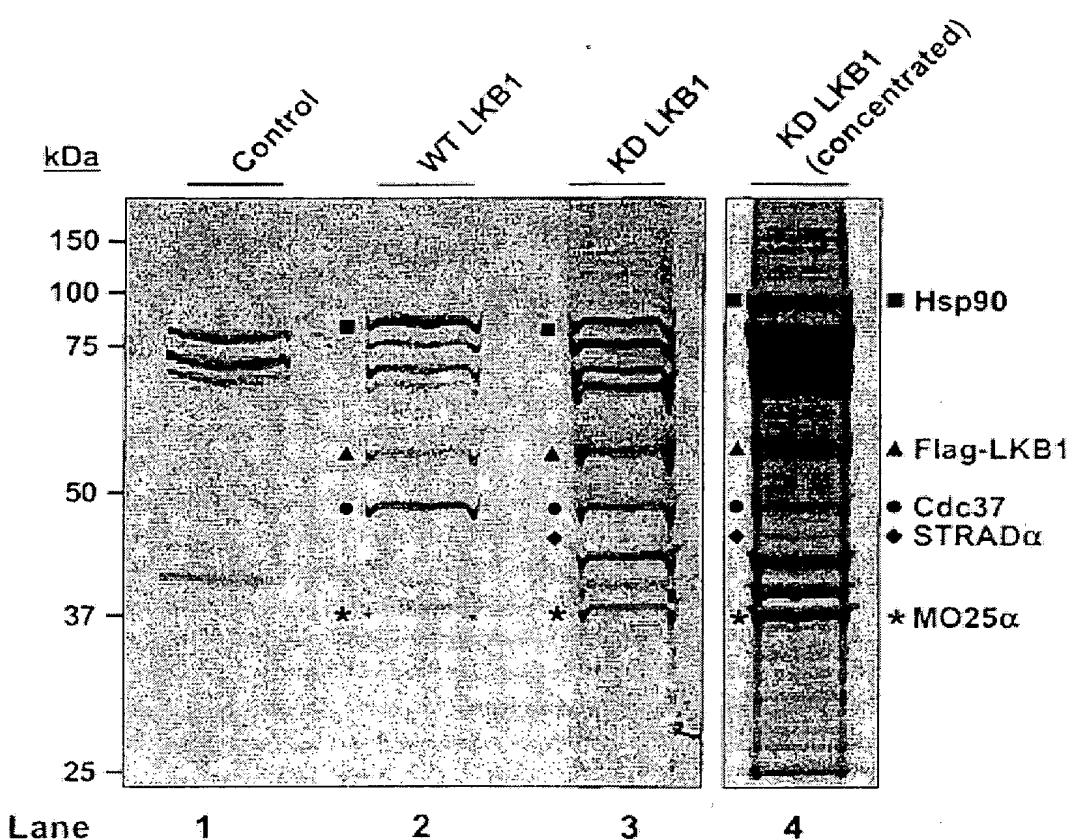
Figure 1A

Figure 1B

Protein name	Peptide matches	% sequence coverage	NCBI gi number
■ Hsp90	15/44	30%	20149594
▲ Flag-LKB1	14/46	35%	7106425
● Cdc37	31/72	59%	5901922
◆ STRAD α	11/80	34%	12060855
* MO25 α	17/37	47%	7706481

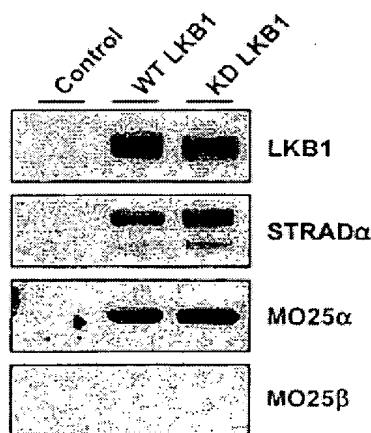
Figure 1C

Figure 2A

(SEQ ID NO:11) hMO25 α	1 MPF-PFGKSHKSPADIVKVNLRKSRAILEKQD-----ISDKKAEKATEEVSKNLVA
(SEQ ID NO:12) hMO25 β	1 MPL-FGKSHKNPADIVKVNLRKSRAILEKQD-----DKKTEKASEEVSKSLOA
(SEQ ID NO:15) dMO25	1 MPL-FGKSCKSPVREIVKSLKERAIALEKAGD-----DKVEKAQEEVSKNLVS
(SEQ ID NO:13) cMO25 α	1 MLKPFLFGKSHKSPADIVKVNLRKSRAILEKAGD-----TNTSFKVEKAIEETAKMLAL
(SEQ ID NO:14) cMO25 β	1 MPL-LFGKSHKSPADIVKVNLRKSRAILEKAGD-----SDKKYDKALEEVSKNLAM
(SEQ ID NO:11) hMO25 α	50 MKEPILYGH--NEKEP-QTEAVAQALAQELYNSGLSTLIAIDLQLIDFEGKKDVQIFNNEL
(SEQ ID NO:12) hMO25 β	51 MKEPILCQH--NEKEP-PTEAVAQALAQELYNSGLLIVLTLIADLQLIDFEGKKDVQIFNNEL
(SEQ ID NO:15) dMO25	52 MKEPILYGH--SEAEPADYVVAQALAQELYNSNLILJLIONLHEIDFEGKKHVALIFNNEL
(SEQ ID NO:13) cMO25 α	53 AKTFPEYGEDANPPNN--EQVTLQAEVYNVWLPPLIKHLHGFPEECKKDVASEFNNEL
(SEQ ID NO:14) cMO25 β	54 MKEPILYGH--SEAEPADYVVAQALAQELYNSNLILPMLIKMLPKFEFECKKDVAQIFNNEL
(SEQ ID NO:11) hMO25 α	107 RROIGTRSPPTVEYICTOONILSMLLKGYE--SPEIALNCGMLRECIRHEPLAKIILIS
(SEQ ID NO:12) hMO25 β	108 RROIGTRSPPTVEYISAPPEILMALKGYE--APQIALRCGMLRECIRHEPLAKIILIS
(SEQ ID NO:15) dMO25	109 RROIGTRSPPTVEYICTPEILEFLIMLGYEDAHPEIALNCGMLRECARYBALAKIILIS
(SEQ ID NO:13) cMO25 α	110 RROIGTRSPPTVEYAPPEILITLLKGYE--QPIALNCGMLRECIRHEPLAKIILIS
(SEQ ID NO:14) cMO25 β	111 RROIGTRSPPTVEYICRPEILEQLEQYS--VPPIALNCGMLRESIRHHLAKIILIS
(SEQ ID NO:11) hMO25 α	165 QPRDFPEVEMSTFDIASDAFTFKDILLTRHKHESAEFFLCHYDEF-SEYERLLHSEN
(SEQ ID NO:12) hMO25 β	166 QPRDFPEVEMSTFDIASDAFTFKDILLTRHKHESAEFFLCHYDEF-SEYERLLHSEN
(SEQ ID NO:15) dMO25	167 QPRDFPEVEMSTFDIASDAFTFKDILLTRHKHESAEFFLCHYDEF-SEYERLLHSEN
(SEQ ID NO:13) cMO25 α	168 YFQRPFVEMESDVFDIASDAPSTFKDILLTRHKHESAEFFLCHYDEF-SEYERLLHSEN
(SEQ ID NO:14) cMO25 β	169 YFNTFFLYVSEVFDDIASDAPSTFKDILLTRHKHESAEFFLCHYDEF-SEYERLLHSEN
(SEQ ID NO:11) hMO25 α	224 VTRROSILKLLGEPLLDRHNFTMTKYISKPENLKLMNNLLRDKSRNIQFEAFHVFKVFVA
(SEQ ID NO:12) hMO25 β	225 VTRROSILKLLGEPLLDRHNFTMTKYISKPENLKLMNNLLRDKSRNIQFEAFHVFKVFVA
(SEQ ID NO:15) dMO25	226 VTRRQSLKLLGEPLLDRHNFTMTKYISKPENLKLMNNLLRDKSRNIQFEAFHVFKVFVA
(SEQ ID NO:13) cMO25 α	227 VTRRQSLKLLGEPLLDRHNFTMTKYISKPENLKLMNNLLRDKSRNIQFEAFHVFKVFVA
(SEQ ID NO:14) cMO25 β	228 VTRROSILKLLGEPLLDRHNFTMTKYISKPENLKLMNNLLRDKSRNIQFEAFHVFKVFVA
(SEQ ID NO:11) hMO25 α	284 NPNKTOPIEDILLRNQAKLKEFPLSKFENDRTDEQFNDEKYLKQIPELKRPAAQEA-
(SEQ ID NO:12) hMO25 β	285 NPNKTOPIEDILLRNQAKLKEFPLSKFENDRTDEQFNDEKYLKQIPELKRPAAQEA-
(SEQ ID NO:15) dMO25	286 NPNKTOPIEDILLRNQAKLKEFPLSKFENDRTDEQFNDEKYLKQIPELKRPAAQEA-
(SEQ ID NO:13) cMO25 α	287 NPNKTOPIEDILLRNQAKLKEFPLSKFENDRTDEQFNDEKYLKQIPELKRPAAQEA-
(SEQ ID NO:14) cMO25 β	288 NPNKTOPIEDILLRNQAKLKEFPLSKFENDRTDEQFNDEKYLKQIPELKRPAAQEA-
(SEQ ID NO:11) hMO25 α	357 KSKEDENQEPAGPSBGPSTSQ-----
(SEQ ID NO:12) hMO25 β	-----
(SEQ ID NO:15) dMO25	-----
(SEQ ID NO:13) cMO25 α	-----
(SEQ ID NO:14) cMO25 β	-----

Figure 2B

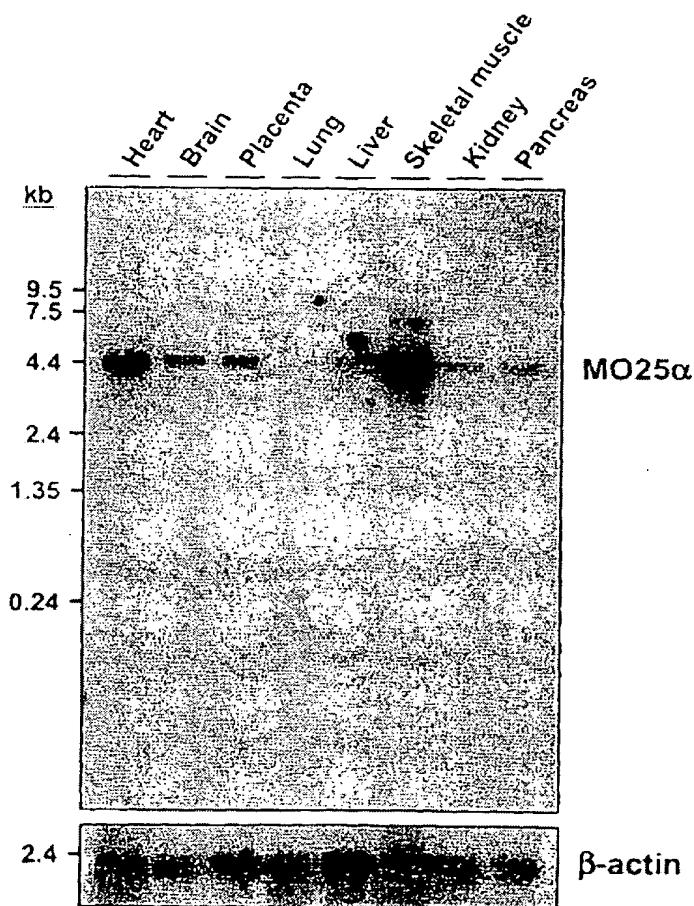


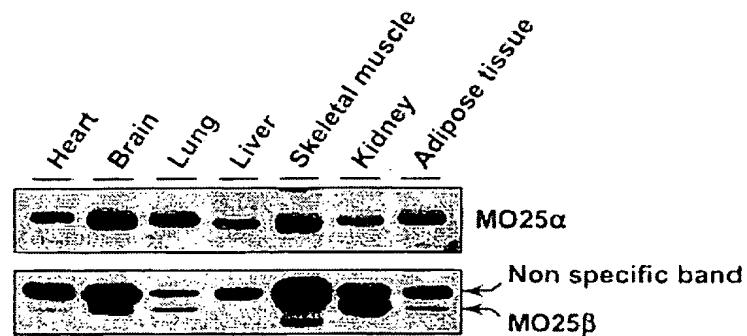
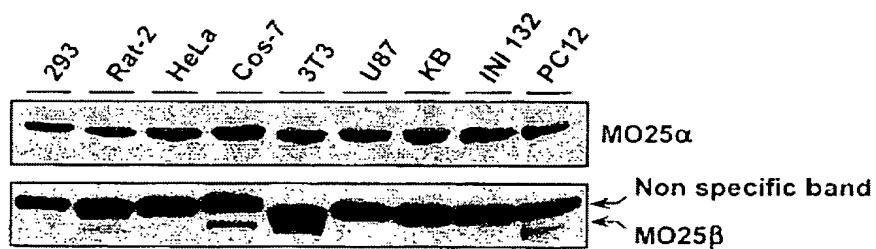
Figure 2C**Figure 2D**

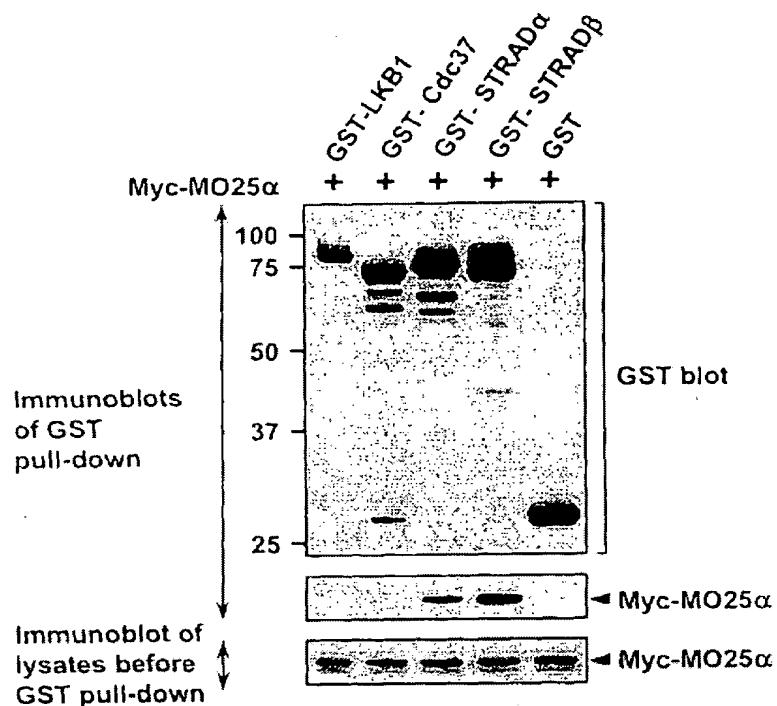
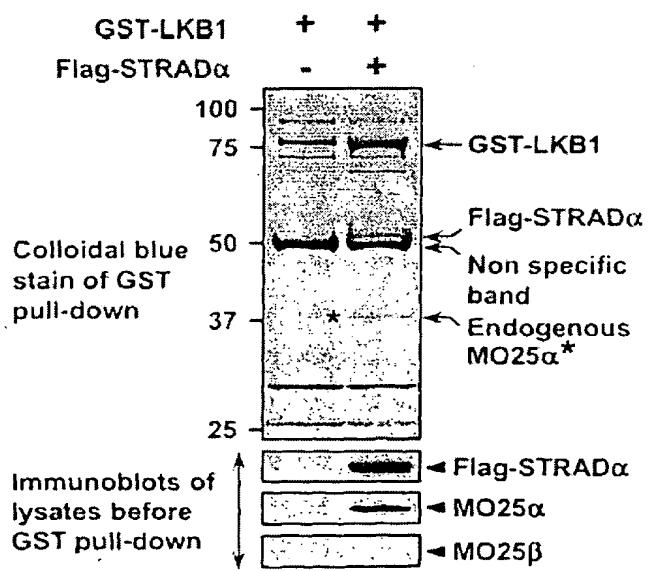
Figure 4A**Figure 4B**

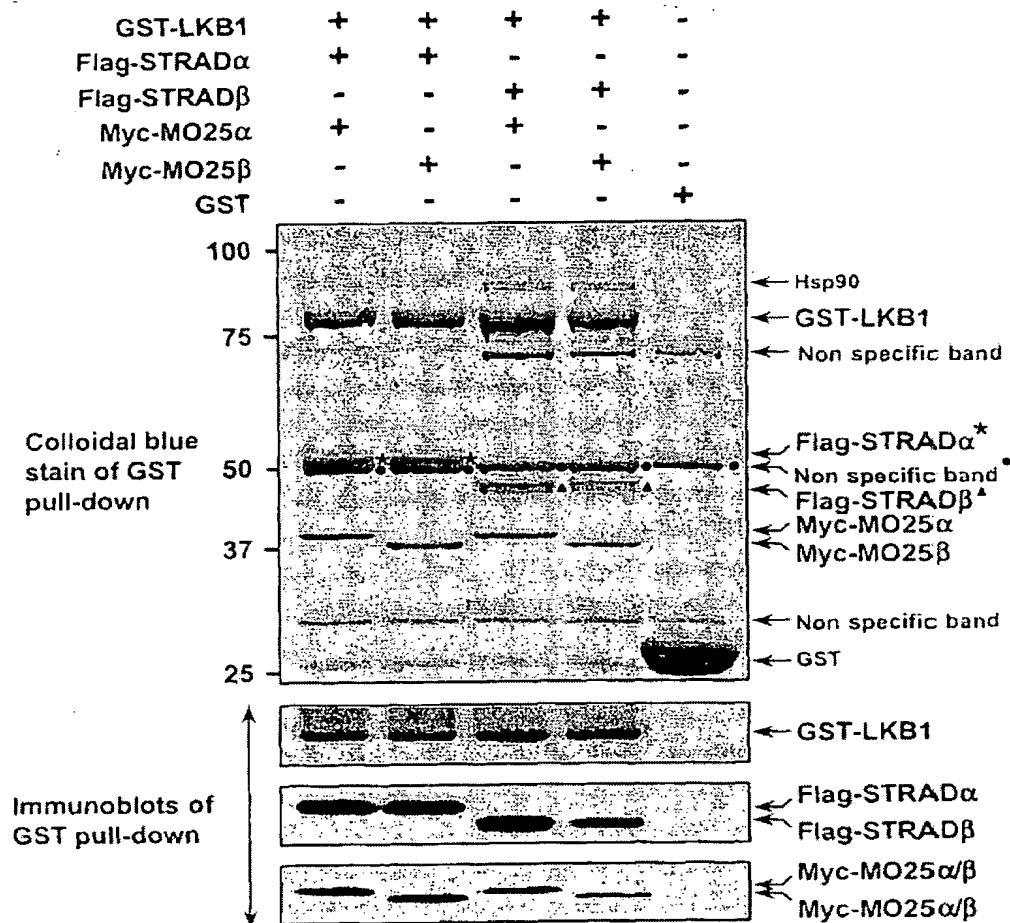
Figure 4C

Figure 5 (continued)

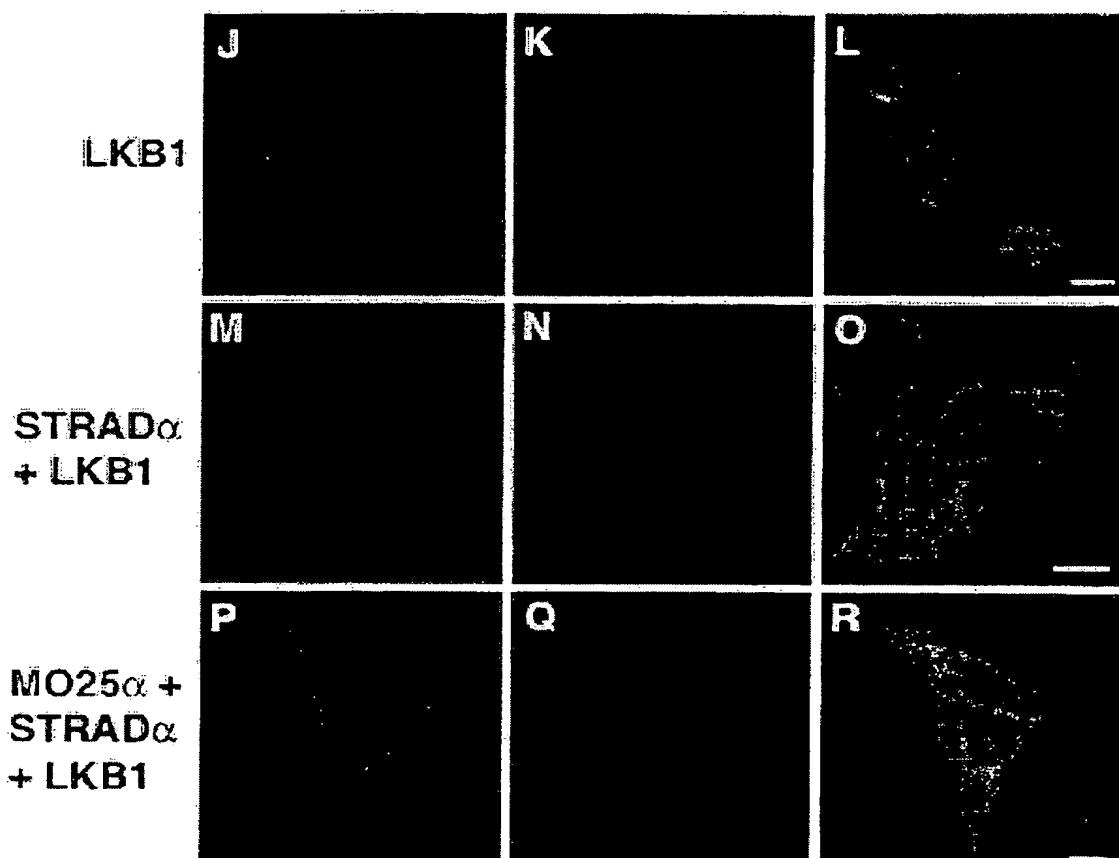


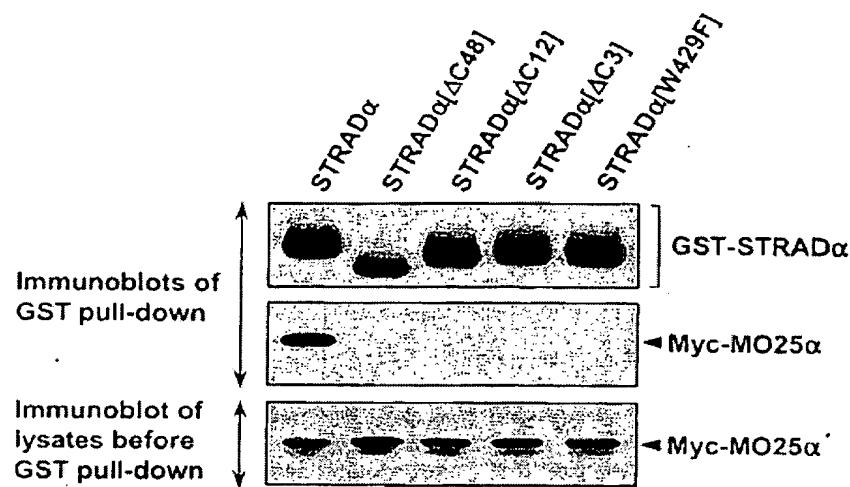
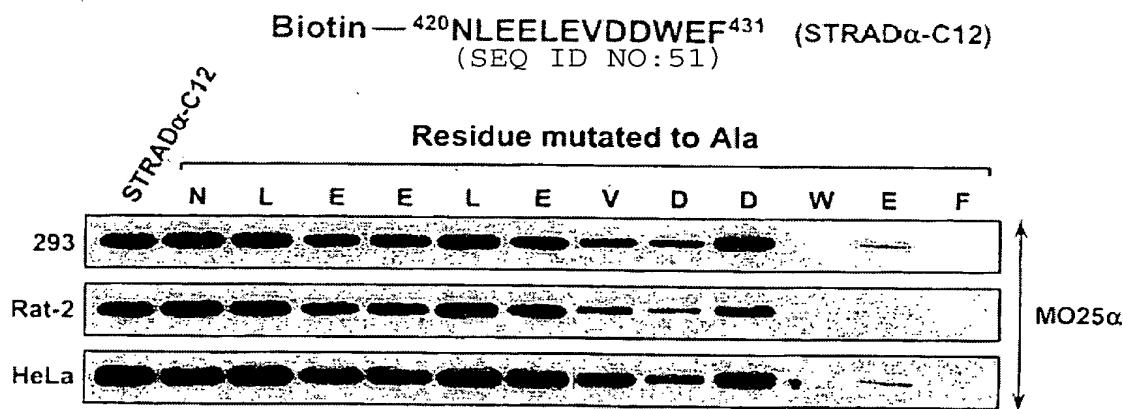
Figure 6A**Figure 6B**

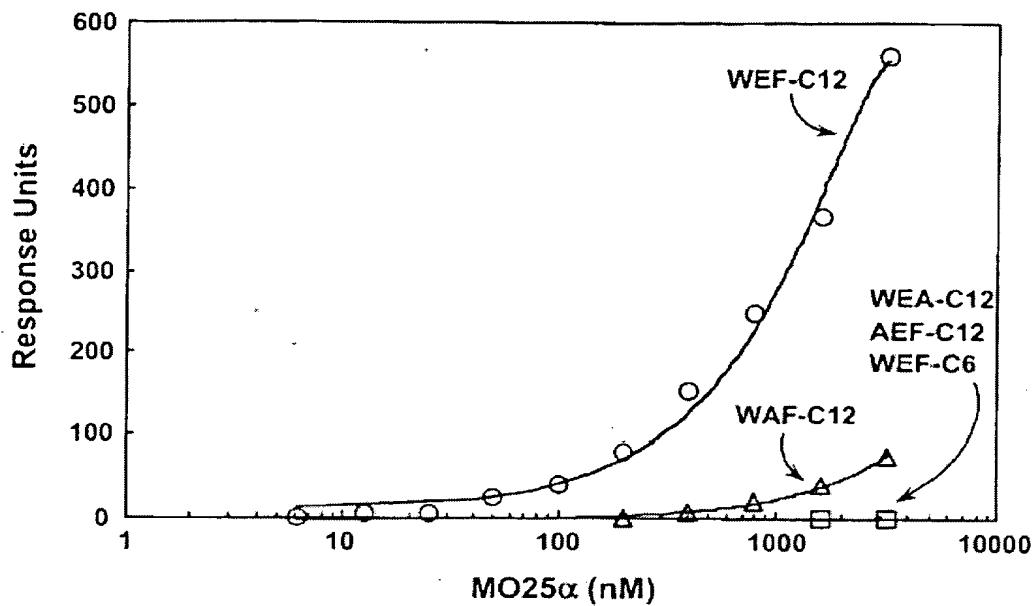
Figure 6C

Figure 10

Figure 12

(SEQ ID NO:58) Tos3 11 .LPRSSLLYNNASNSSRIKETRKVKLLYNPLTKR.....Q...ILNNFEILATLGNGQ
 (SEQ ID NO:59) Pak1 94 ..PTTTTSSFCSSGSSKNVKETNRISLTYPVSKR.....K...VLNTYEIIKELGHGQ
 (SEQ ID NO:61) CaMKK β 121 CICPSLPYSPVSSPQSSPRLPRRPTVESHHSITGM.....QDCVQLNQYTLKDEIGKGS
 (SEQ ID NO:6) LKB1 7 QQLGMFTEGELMSVGMDTFIHRIDSTEVYQP..RR.....KRAKLIGKYLMGDLLGEQS
 (SEQ ID NO:60) Elml 39 TSSFGSSFSQQKPTYSTIIGENIHTILDEIRPYVKKITVSDQDKKTINQYTLGVSGSQQ
 (SEQ ID NO:62) consensus 121 p ss s s ss rik tv l y pltkr q ilnny i 1G Gq

(SEQ ID NO:58) Tos3 61 YGKVKLARDLGTGALVAIKILNRFKEKRS....GYSL.....QLKV.EN.....
 (SEQ ID NO:59) Pak1 144 HGKVKLARDILSKQLVAIKIVDRHEKKQRKFFTFIK.....SSKISEN.....
 (SEQ ID NO:61) CaMKK β 176 YGVVKLAYNENDNTYYAMKVLSK..KKLIRQAGFPR.....RPPPRGTRPAPGGCIQP
 (SEQ ID NO:6) LKB1 60 YGKVKEVLDSETLCRRRAVKILKK..KKLRR.....I
 (SEQ ID NO:60) Elml 99 FGYVRKAYSSTLGKVVAVKIIPKKPWNAAQYQSVNQVMRQIQLWKSKGKITTNMSGNEAMR
 (SEQ ID NO:62) consensus 181 yGkVla d t lvAiKil k kk k y k

(SEQ ID NO:58) Tos3 99PRVNQEIEVMKRCHHE.NVVELYEILNDPESTKVLVLEYCSRGPVKWCPEKMEI
 (SEQ ID NO:59) Pak1 187DKIKREIAIMKKCHHK.HVVKLVEVLDLKSRIYLVLEYCSRGEVKWCPPDCMES
 (SEQ ID NO:61) CaMKK β 227 RGPIEQVYQEIAILKKLDHP.NVVKLVEVLDPNEDHLYMVFEVLVNOGPV.....MEV
 (SEQ ID NO:6) LKB1 89 PNGEANVKKEIQLLRLRHK.NVIQLVDVLYNEEKQKMYMMVMEYC.....VCGMQEM.L
 (SEQ ID NO:60) Elml 159 LMNIEKCRWEIFAAASRLRNNHVIRVLIECLDSPFSESIWIVTNWCSLGELOWKRDDEDI
 (SEQ ID NO:62) consensus 241 drvk EI vmkrlhh nvv LievLddp s kvylVleycs g v wc mei

(SEQ ID NO:58) Tos3 154 .KAVGPSILTFOQ....SRKVLDVVSGLEYLHSQGITHRDIKPSNLLISSNGTV.KISD
 (SEQ ID NO:59) Pak1 242 .DAKGPSLLSFQE....TREILRGVVLGLEYLHYQGIIHRDIK PANLLISGDGTv.KISD
 (SEQ ID NO:61) CaMKK β 279 .PTLKP..LSEDQ....ARFYFQDLIKGIEYLHYQKTIHRDIKPSNLLVGEDGHI.KIAD
 (SEQ ID NO:6) LKB1 141 .DSVPEKRFPVQC....AMGYFCQLIDGLEYLHSQGIVHKDIKPGNLLTTGGTL.KISD
 (SEQ ID NO:60) Elml 219 LPQWKKIVISNCSTFAKKILEDMTKGLEYLHSQGCIHRDIKPSNILLDEEEVAKLSD
 (SEQ ID NO:62) consensus 301 v p ils q ar vv dvv GLEYLHsQgiiMrDIKPsNLLis dgcv KisD

(SEQ ID NO:58) Tos3 208 FG..VAM.STATGSTNIQSSHEQLLKSRALGTPAFFAPELCSTEKEY.....
 (SEQ ID NO:59) Pak1 296 FG..VSLAASSTNSSDSSESLELAKTVGTPAFFAPEMCLGEDAFTRYNLTKENLFRG
 (SEQ ID NO:61) CaMKK β 331 FG..V.....SNEFKGS..DALLSNVTGTPAFMAPESLS.....ETRKIFSG
 (SEQ ID NO:6) LKB1 195 LG..VAAEALHPFAADDTCRTSQ.....GSPAFQPPEIANGLDTFS.....
 (SEQ ID NO:60) Elml 279 FGSCIFT PQSLPFSANFEDCFQRELNKIVGTPAFIAPELCHLGNSKRDFVTD.....
 (SEQ ID NO:62) consensus 361 fG v t s d s l r vGtPAF aPElc y

(SEQ ID NO:58) Tos3 252 SC.SSAIDIWSLGVTIYCLLFGKLPFNANSGLELFDSTIINKPLEFPSYEEMLNGATSGIT
 (SEQ ID NO:59) Pak1 354 SCISFMIDIWAVGVTLYCLLFGMLPFFSDFELKLFKIVNDPLKFPTFKEIQSNKVKVSKVS
 (SEQ ID NO:61) CaMKK β 369 K....ALDVWAMGVTLFCVFGQCPFMDERIMCLHSKIKSQALEFPDQPDIA.....
 (SEQ ID NO:6) LKB1 233 ...GFKVDIWSAGVTLYNITTGLYPFEGDNIYKLFENIGKGSYAIP.....
 (SEQ ID NO:60) Elml 332 ...GFKLDIWSLGVTLCLLYNELPFFGENEFETYHKIIIEVSLS SKINGNTLNDLVIKRL
 (SEQ ID NO:62) consensus 421 f idIWs1GVT1Ycllfg 1PF ad 1 lfdki 1 fp em

(SEQ ID NO:58) Tos3 311 M.EEYT...DAKDLLKKLLQKDPDKRIKLA DIKVHPFMC....HYGKSDAASVL...TN
 (SEQ ID NO:59) Pak1 414 CEEEYE...MAKDLLLKLLKLEKNPQKRMTI PAIKKHFV S.WDFDHVPENDEKLLS...SV
 (SEQ ID NO:61) CaMKK β 417E...DLKDLITRMLDKNPESRIVVPEIKLHFWVTRHGAPEPLPSEDENCTLVEVTE
 (SEQ ID NO:6) LKB1 276 .GDCGP...PLSDLKGMLYEPAKRF SIRQIRQHSWFRK..KHPPAEAPVPIPSSPDT
 (SEQ ID NO:60) Elml 389 LEKDVTLRISIYQDLVKVL SRDQPIDSRNHSQISSSS.VNPVNR EGPVRRFFGRLLTKKGK
 (SEQ ID NO:62) consensus 481 ee . 1kDLkkleknP kri l 1k hpfv dh p d v1 t

(SEQ ID NO:58) Tos3 359 LETFHELKVSP...SSCKRVELVSLPVNSSFASLD SVY MENFDHNNLRTGADRNS
 (SEQ ID NO:59) Pak1 467 LE..QKLRF.....QCNQTDQFE.PISISKHELKN AV.....SGVGKKIKESV
 (SEQ ID NO:61) CaMKK β 469 EEEVENSVKHIPS LATVILVKTMIRKRSFGNP FEGS RREERLSA PGNLLTKQGSEDNLQG
 (SEQ ID NO:6) LKB1 329 KDRWRSM TVV PYLEDLHG ADEDED LF DIEDDIIY TDFTVPGQVPEEEASHNGQRRGLPK
 (SEQ ID NO:60) Elml 448 KKTSGKGDKVVLVSATSKVTPSIHIDE PDKECFSTVLRSSPDSSDYCSSLGE EAIQVT
 (SEQ ID NO:62) consensus 541 e 1k p l rve pv s 1ks s lg

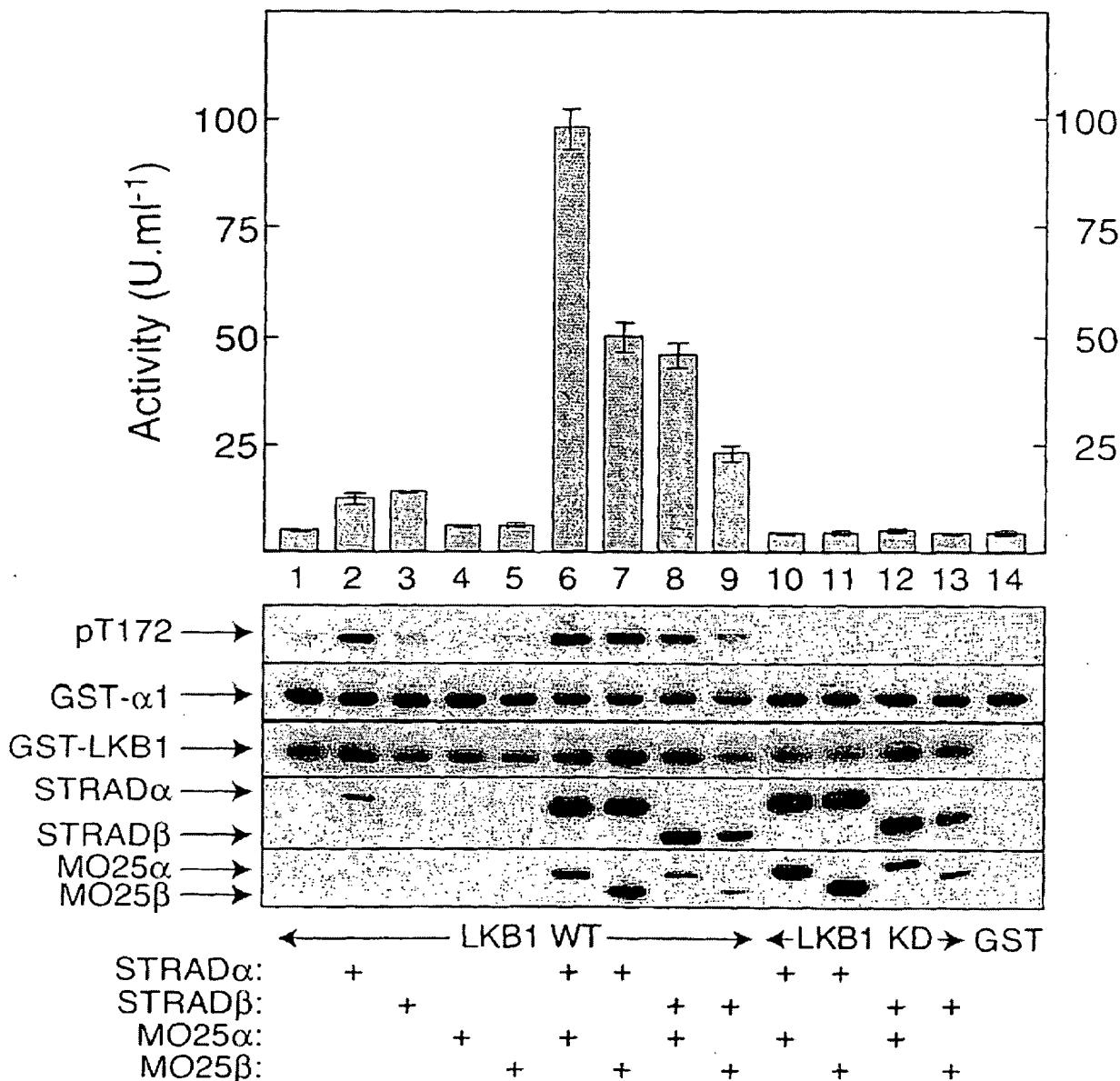
Figure 15AActivation of AMPK α 1 catalytic domain by LKB1

Figure 15B

Phosphorylation of AMPK α 1 catalytic domain by LKB1

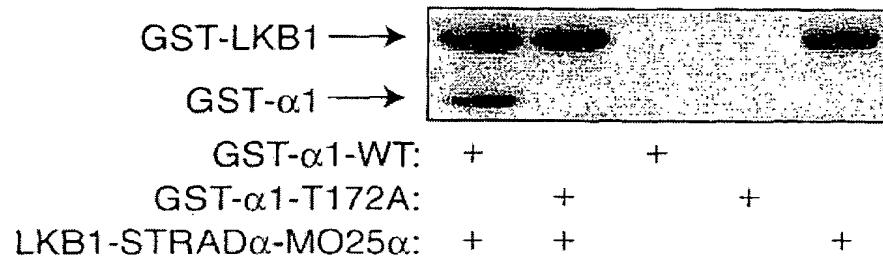
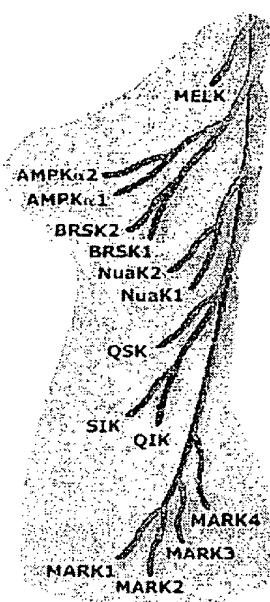


Figure 19

↓¹¹
 ↓¹² ↓¹⁰⁻⁹ ↓⁻⁵ ↓^{-3 -2} P
 ↓ ↓ ↓ ↓

(SEQ ID NO:63) PKA-C α	239 DFGFAKR..V.KG.RTWTLCGTPEYLAPE
(SEQ ID NO:64) PKC α	539 DFGMCKEHMM.DGVTRTRFCGTPDYIAPE
(SEQ ID NO:65) NuaK1	196 DFGLSNLYQKDK..FLQTFCGSPLYASPE
(SEQ ID NO:66) NuaK2	237 DFGLSNLYHQGK..FLQTFCGSPLYASPE
(SEQ ID NO:67) BrsK1	190 DFGMASLQVGDS..LLETSCGSPHYACPE
(SEQ ID NO:68) BrsK2	159 DFGMASLQVGDS..LLETSCGSPHYACPE
(SEQ ID NO:69) SIK	167 DFGFGNFYKSGE..PLSTWCGSPPYAAPE
(SEQ ID NO:70) QIK	160 DFGFGNFFKSGE..LLATWCGSPPYAAPE
(SEQ ID NO:71) AtSnRK1- α 1	160 DFGLSNIMRDGH..FLKTSCGSPNYAAPE
(SEQ ID NO:72) AtSnRK1- α 2	161 DFGLSNVMRDGH..FLKTSCGSPNYAAPE
(SEQ ID NO:73) AMPK- α 1	159 DFGLSNMMSDGE..FLRTSCGSPNYAAPE
(SEQ ID NO:74) AMPK- α 2	157 DFGLSNMMSDGE..FLRTSCGSPNYAAPE
(SEQ ID NO:75) ScSnf1	195 DFGLSNIMTDGN..FLKTSCGSPNYAAPE
(SEQ ID NO:76) QSK	206 DFGFSNLFTPQ..LLKTWCGSPPYAAPE
(SEQ ID NO:77) MELK	150 DFGLCAKPKGNKDYLQTCGSLAYAAPE
(SEQ ID NO:78) consensus	243 DFGlsnl g fL TsCGSp YAAPE

Figure 21A

(SEQ ID NO:73)	AMPK1	159-	DFGLSNMMSDCE--FLRTSCGSPNYAAPE	*	*
(SEQ ID NO:74)	AMPK2	157-	DFGLSNMMSDCE--FLRTSCGSPNYAAPE		
(SEQ ID NO:67)	BRSK1	174-	DFGMASIVQVGDS--LLETSCGSPHYACPE		
(SEQ ID NO:68)	BRSK2	159-	DFGMASIVQVGDS--LLETSCGSPHYACPE		
(SEQ ID NO:65)	NUAK1	196-	DFGLSNIVYQKDK--FLQTTECGSPLVASPE		
(SEQ ID NO:66)	NUAK2	193-	DFGLSNIVHOGK--FLQTTECGSPLVASPE		
(SEQ ID NO:69)	SIK	167-	DFGFGNFYKSGE--PLSTWCGSPPYAAPE		
(SEQ ID NO:70)	QIK	160-	DFGFGNFEKSGE--LLATWCGSPPYAAPE		
(SEQ ID NO:76)	QSK	206-	DFGFSNIEIPGQ--LLKTWCGSPPYAAPE		
(SEQ ID NO:79)	MARK1	200-	DFGFSNEEVVGN--KLDTECGSPPYAAPE		
(SEQ ID NO:80)	MARK2	160-	DFGFSNEEVVGN--KLDTECGSPPYAAPE		
(SEQ ID NO:81)	MARK3	196-	DFGFSNEEVVGG--KLDTECGSPPYAAPE		
(SEQ ID NO:82)	MARK4	198-	DFGFSNEEVILGS--KLDTECGSPPYAAPE		
(SEQ ID NO:77)	MELK	150-	DFGLCAKPKGNKDYLQTCCGSLAYAAPE		

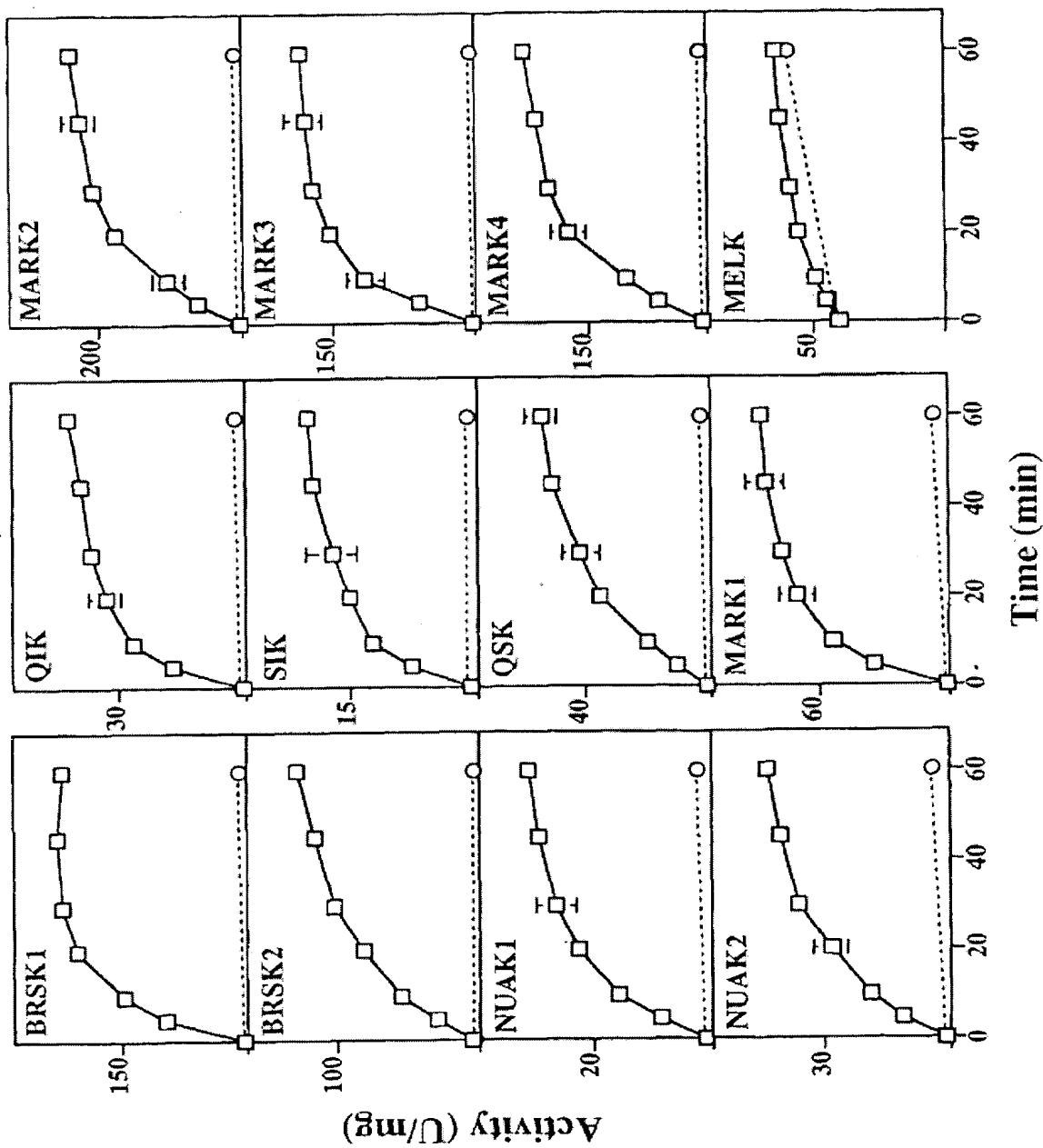
Figure 21B

Figure 26A

	Peptide	K_m (nM)	V_{max} (U/mg)	V_{max}/K_m
NUAK2	LSNLHYQGKFLQTFCGSPLYRR (SEQ ID NO:83)	0.15 ± 0.02	86.8 ± 3.4	578
SIK	FGNFYKSGEPLSTWCGSPPYRR (SEQ ID NO:84)	0.46 ± 0.05	63.9 ± 2.2	139
AMPK	LSNMMSDGEFLR _T SCGSPNYRR (SEQ ID NO:85)	1.40 ± 0.24	94.1 ± 8.3	67
BRSK2	MASLQVGDSLE _T SCGSPHYRR (SEQ ID NO:86)	0.40 ± 0.06	10.6 ± 0.6	26
MARK3	FSNEEFTVGGKLDT _T FCGSPPYRR (SEQ ID NO:87)	0.62 ± 0.06	10.2 ± 0.4	16
MELK	AKPKGNKDYHLO _T CCGSLAYRR (SEQ ID NO:88)	>3	>60	

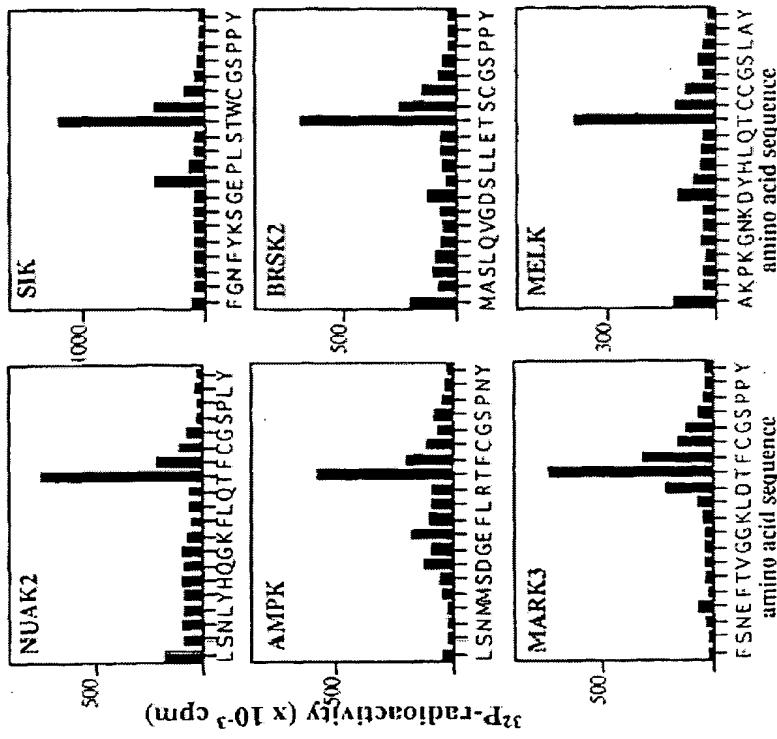
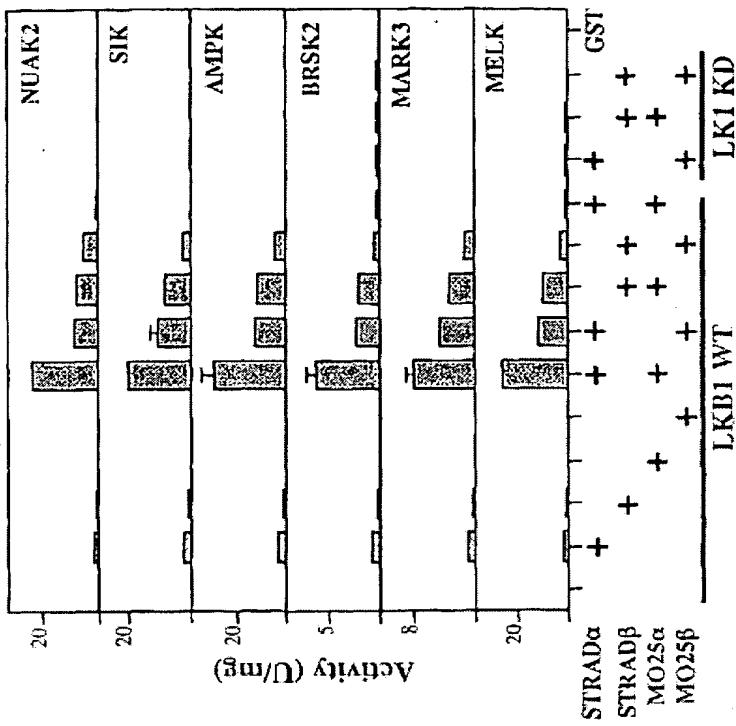
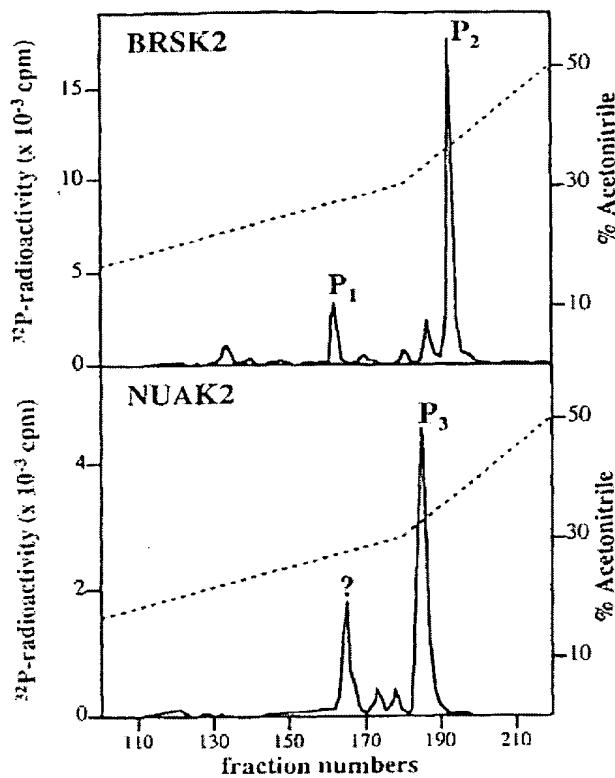
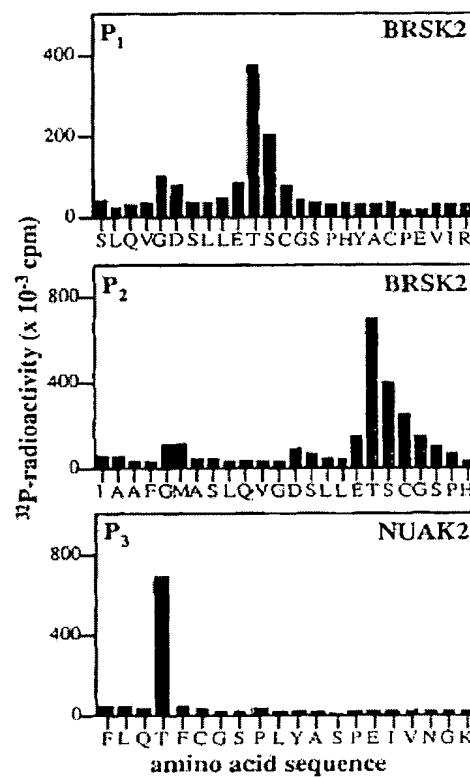
Figure 26B**Figure 26C**

Figure 30A**Figure 30B****Figure 30C**

Kinase	Phosphopeptide	Mass observed	Theoretical mass
BRSK2 P_1	IAAFGmASLQVGDSLLET(p)SCGSPHYACPEVIR (SEQ ID NO:89)	3268.7870	3628.6680
BRSK2 P_2	SLQVGDSLLET(p)SCGSPHYACPEVIR (SEQ ID NO:90)	2951.4530	2951.3472
NUAK2 P_3	FLQT(p)FCGSPLYASPEIVNGK (SEQ ID NO:91)	2356.1088	2356.1333
MARK4	LDT(p)FCGSPPYAAPELFQGK (SEQ ID NO:92)	2225.9983	2226.1497
MELK	GNKDYHLQT(p)CCGSLAYAAPELIQCK (SEQ ID NO:93)	2970.4421	2970.3648